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The application of *Vicia ervilia* protein isolate - *Launaea acanthodes* gum nanocomposite film containing *Silybum marianum* extract microcapsule and graphene oxide nanoparticle for packing Koupeh cheese

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ABSTRACT

Cheese is known as the most challenging dairy product due to its characteristics and diversity, which makes edible coatings and films widely applied to extend its shelf life. Therefore, this research aims to application of *Vicia ervilia* protein isolate - *Launaea acanthodes* gum nanocomposite film containing *Silybum marianum* extract microcapsule at different levels (0 and 15 V/V) and graphene oxide nanoparticle at different levels (0 and 4 W/V) for packing Koupeh cheese was done . Two storage time factors and samples of produced films (control film, film containing maximum amounts of nanoparticles and *Silybum marianum* extract microcapsule and optimal film containing 3.41% W/V of nanoparticles and 11.35% V/V of *Silybum marianum* extract microcapsule) in the packaging of Koupeh cheese and Koupeh cheese samples without film coating, were investigated according to the response surface method of the factorial design. Physicochemical characteristics (pH, acidity, salt, fat, protein), microbial (total count of microorganisms, mold and yeast) and sensory evaluation of cheese during 60 days of storage were investigated. The effect of film packaging on coupe cheese during the storage period caused a decrease in pH and fat, an increase in acidity and salt. Also, the results showed that the increase in storage time and type of packaging had no significant effect on the amount of protein and sensory evaluation of cheese samples (P>0.05). The microbial characteristics of cheese during 60 days of storage were within the permissible limits of the national standard. In general, *Vicia ervilia* protein isolate - *Launaea acanthodes* gum nanocomposite films containing *Silybum marianum* extract microcapsule and graphene oxide nanoparticles may be suitable for use as environmentally friendly packaging materials in the food industry and also increase the safety of food products during storage.

1.Introduction

High oxygen concentrations promote microbial growth and oxidation of dairy products. Exposure to light can also lead to discoloration, loss of nutrients, and off-flavors, and the presence of moisture can cause dairy products to spoil much more quickly. Cheese is known as the most challenging dairy product due to its many characteristics and variations, which has led to the widespread use of edible coatings and films to extend its shelf life. Packaging materials used for dairy products, and especially cheese, must be fully permeable to carbon dioxide to extend its shelf life and prevent the growth of microorganisms [1]. Koupeh cheese is a traditional cheese with a semi-hard, creamy texture, high fat content, and a sharp flavor and aroma, and is known as Pot cheese or Juge cheese. The largest production of this type of cheese is in Greece, Turkey, and in the northwest of Iran, where it is known as Kouzeh cheese or Koupeh cheese [2-4, 5].

Vicia ervilia, belonging to the legume family and within the genus *Vicia*, is a part of a group of 160 species. This particular species is cultivated across approximately 540,761 hectares in temperate regions of Europe, West and Central Asia, North Africa, and America [6, 7]. One of the annual species within the Vicia genus is known by its scientific name *V. ervilia*. This plant is commonly cultivated in Mediterranean regions due to its use as forage, its ability to cover saline soils, and its high nutritional value. Additionally, it is grown widely due to its resistance to drought and insects, ease of cultivation and harvesting, its independence from fertilizers, and the presence of nitrogen-fixing bacteria in its roots [6, 8]. *V. ervilia* contains approximately 26% protein, with some sources reporting as high as 32%. This protein content is nearly double that of cereals. It also comprises 1.6% fat, 61.2% carbohydrates, 5.9% crude fiber, and 3.7% ash[6]. The primary amino acids present in *V. ervilia* include glutamic and aspartic acids, arginine, and leucine. However, this source lacks methionine and cysteine. Generally, the amino acid profile of *V. ervilia* is similar to lysine-rich soy flour [9]. Therefore, due to its high protein content, *V. ervilia* is an economically viable source for producing edible films with functional and suitable appearance properties for food packaging applications [10].

Launaea acanthodes gum (LAG) is an annual plant and belongs to the Lactuceae tribe in the *Asteraceae* (*Compositae*) family. Upon cutting or causing physical damage to the lower branches of this plant, it exudes a white, sour latex that transforms upon exposure to air into a non-transparent yellow substance, possessing a pungent taste and a waxy appearance [11]. This plant is also known by various names such as "Melk Azraq," "Charkhan," and "Charkheh" and features stemless and branchless bushy growth. It thrives in arid regions, particularly in central (desert) areas [12]. The secretion from this plant contains compounds with low and relatively high molecular weight, and it is used in treating gastrointestinal disorders, wound healing, and stomach ulcers. The chemical compounds of this plant include terpenoids, alkaloids, flavonoids, and saponins, as well as proteins and polysaccharides [13]. Galactose, rhamnose, arabinose, and residual galacturonic acid constitute monosaccharides in this plant, and arabinogalactan is the main compound in the chain of LAG [11]. Overall, due to its high solubility and viscosity, this gum could possess diverse capabilities in food applications. However, bio-packaging materials currently have some limitations, and they cannot fully match the characteristics of petroleum-based materials [14].

Silybum marianum also recognized as *thistle Milk*, is a medicinal plant classified as *S. marianum* (*L.*) *Gaertn*. it belongs to the *Asteraceae* family and the *Asteraceae* tribe [15, 16]. *S. marianum* typically originates from Mediterranean regions but is growing in warm, arid areas and the western regions of Iran, such as Abadan, Bushehr, Gorgan, and Gonbad. Moreover, it is cultivated in Asian, European, and American countries [17]. *S. marianum* is a biennial or annual thistle plant featuring prickly stems and erect, ranging from 50 to 70 centimeters in height. It bears green leaves with toothed margins, reddish-purple large flowers, and its fruit is black with yellow spots [18]. *S. marianum* is a rich source of phytochemical compounds. Silymarin is the standardized extract of this plant containing three main compounds: silybin, silydianin, and silychristin, among which silybin exhibits the

highest biological activity. Other flavonolignans such as isosilybin, silyandrin, silyhermin, and neosilyhermin are also present [19]. The seeds of this plant contain the highest percentage of silymarin, a potent antioxidant. Therefore, *S. marianum* seeds are utilized as a rich source of antioxidants for substituting synthetic antioxidants and food preservatives due to their free radical scavenging activity against DPPH and their prevention of lipid peroxidation [20]. So to preserve *S. marianum* sensitive and desirable compounds and countering the high volatility of the extract, *S. marianum* extract (SME) is encapsulated via different methods that can lead to the preservation and manufacturing of new materials with novel features [21].

Graphene oxide nanoparticles (GONPs) have gained attention for their ability to enhance the mechanical-thermal properties and chemical stability, thereby improving the characteristics of films due to their high surface area-to-volume ratio [22]. Graphene oxide, due to the presence of hydroxyl, carboxyl, and epoxy functional groups on its surface, is a hydrophilic compound capable of forming hydrogen bonds and adsorbing metals or other mineral precursors [22, 23]. Moreover, graphene oxide is one of the best reinforcing agents in polymer composites due to its antimicrobial activity against Gram-positive and Gram-negative bacteria [24]. Furthermore, due to its biocompatibility and unique physicochemical properties, this nanoparticle has been employed in biological applications such as drug delivery, tissue engineering, and glucose sensing [25]. So far, no subject has been worked on under the title of *Vicia ervilia* protein isolate/*Launaea acanthodes* gum nanocomposite film containing *S. marianum* extract microcapsules (SMEM) and GONPs, and this is the innovation of the article. The hypothesis of this research is that the film containing microcapsules of SME and GONPs leads to an increase in the shelf life of Koupeh cheese. The aim of this research is to use a *vicia ervilia* protein isolate - *Launaea acanthodes* gum nanocomposite film containing microcapsules of *Silybum marianum* extract and graphene oxide nanoparticles for packaging Koupeh cheese.

2. Materials and Methods

2.1. Materials

The seeds of *V. ervilia* and *S. marianum* were purchased from the Urmia market in Iran, while LAG was purchased from Tehran, Iran. GONPs were purchased from the US-Nano company in the United States, with a purity of 99% and a particle size ranging from 3.7 to 4 nm. Wheat starch was acquired from the Merck company in Germany, Koupeh cheese from Pegah Factory, Urmia (pH 4.62, acidity=0.8% by weight lactic acid , salt=3.6%, fat=25%, protein=21.2%), and the bacterial strains *Escherichia coli* (ATCC 25922) and *Staphylococcus aureus* (ATCC 25923) (The Collection of Industrial Microorganisms of Iran) and other chemicals were analytical grade and were provided from Merck (Germany).

2.2. Methods

2.2.1. Preparation of nanocomposite film for packaging Koupeh cheese

To produce nanocomposite film, 2 gr of isolated *V. ervilia* protein powder was dissolved in 50 ml of distilled water and the pH was adjusted to 12 with 1 M NaOH and stirred on a magnetic stirrer at 500 rpm for 1 hour without applying temperature; then, 2 gr of LAG gum was weighed and stirred in 50 ml of distilled water for 1 hour at 40-50°C on a magnetic stirrer at 500 rpm; after the gum dissolved, the resulting mixture was centrifuged at 3500×g for 10 minutes to separate impurities and insoluble materials, and then these two polymers were composited together for 1-2 hours; Graphene oxide nanoparticles were added to the film solution in different amounts according to the statistical design and stirred on a magnetic stirrer for 15 minutes, then placed in an ultrasound bath (Model Soner203, Rocker Taiwan) without temperature for 45 minutes, and then the extract microcapsules were added according to the statistical design and stirred again on a magnetic stirrer without temperature for 30 minutes. Then, glycerol was added as a plasticizer at a rate of 40% of the total dry matter weight and stirred for 45 minutes, and finally, 100 ml of the film solution was poured into a disposable container with specific dimensions (11×17 cm) using a pipette and dried in an oven at 34 °C [26-28]. The dried nanocomposite film was used for antimicrobial packaging and storage of Koupeh cheese. For this purpose, approximately 35 gr of Koupeh cheese was prepared, weighed under sterile conditions, and packaged in films containing

different percentages of microcapsules of *Silybum marianum* extract and GO nanoparticles according to the statistical design in Table (1) and stored in disposable containers with lids for 60 days in the refrigerator at 4 ° C, as shown in Figure (1). The relevant tests were performed on the samples during specific storage periods (immediately after packaging, days 15, 30, 40, and 60).

Figure (1): Koupeh cheese samples packed in composite films

2.3. Physicochemical tests of packaged Koupeh cheese samples

2.3.1. Acidity

The cheese sample was thoroughly softened and mixed in a mortar and transferred to a suitable container with a lid, then, 20 gr of the test sample was weighed and dissolved in some distilled water, after adding all the contents, it was made up to volume in a 250 ml volumetric flask. It was filtered using a strainer and 25 ml of the filtered solution was transferred to a suitable beaker. 0.5 ml of phenolphthalein was added to it and titrated with 0.1 normal sodium hydroxide. This process was continued until a pale pink color appeared and the color was stable for 5 seconds. Finally, the acidity of the cheese was calculated in terms of lactic acid using equation (1) as follows [29]:

$$A = \frac{N \times 0.009}{M1} \times 100 \qquad \text{(Eq. 1)}$$

Where: N is the amount of milliliters of 0.1 normal sodium hydroxide used; M_1 is the test weight; A is the percentage of acidity.

2.3.2. pH

In a beaker, 10 gr of shredded cheese was carefully weighed and 50 ml of distilled water was added to it. The sample was completely homogenized and uniform, and then the fat collected on the surface of the sample was removed with a spatula. Then the pH of the cheese was read and recorded using a calibrated pH meter while stirring (the test temperature was about 20°C) [30].

2.3.3. Salt measurement

The Mohr titration method is used to measure the salt content in cheese. According to this method, 10 g of cheese sample is crushed in a

laboratory mortar and some boiled distilled water is added to it, then the suspension is stirred well and the volume is brought to 100 ml. After filtering the contents of the flask using a filter paper, 25 ml of the filtered solution is transferred to an Erlenmeyer flask and 1 ml of 3% potassium chromate indicator is added to it and titrated with 0.1 M silver nitrate solution until it reaches a brick red color [31] and the salt content is calculated using equation (2).

$$\%\text{Salt} = \frac{V \times 0.1 \times 58.5}{5} \quad \text{(Eq. 2)}$$

V Amount of ml of 0.1 normal silver nitrate used.

2.3.4. Protein measurement

The total protein content of Koupeh cheese was measured using the macro-Kjeldahl method. In this method, the amount of nitrogen in the sample is calculated and then the protein content of the cheese sample is determined using the protein factor, which is 6.38 for cheese. 1.2 g of the cheese sample was taken and added to the digestion tube of the Kjeldahl apparatus under the hood along with 2 tablets (about 20 g) of Kjeldahl catalyst and 20 ml of concentrated sulfuric acid. The tube was placed in the digestion apparatus, which had previously reached a temperature of 420 °C, for 30 minutes until the color of the solution completely turned light green. After acid digestion, the tube was removed from the digestion oven and cooled, and then diluted with distilled water. The tube with the digested and diluted sample was placed in the distillation apparatus. An Erlenmeyer flask containing Mittel Red indicator was placed under the outlet of the condenser, then boric acid and sodium hydroxide were automatically introduced into the Erlenmeyer and digestion tube by the device and distillation was performed for 4 minutes. The ammonium borate solution formed was titrated with 0.1 M hydrochloric acid until a grayish-purple color was formed and then the protein content in the cheese sample was determined using the titrated value and equations (3 and 4) [32].

$$\%\text{Nitrogen in cheese} = \frac{0.14 \times A}{M} \quad \text{(Eq. 3)}$$

$$\%\text{Protein in the sample} = \%\text{Nitrogen} \times 6.38 \quad \text{(Eq. 4)}$$

2.3.5. Fat measurement

A Soxhlet apparatus (Universal Extractor E-800 model) was used to measure fat. For this purpose, 5 gr of cheese sample was first dried in an oven and then ground in a mortar. Defatting was performed in a Soxhlet apparatus for 4 hours using n-hexane solvent [33], and then the fat content was calculated using equation (5).

$$\%\text{Fat} = \frac{X2 - X1}{M} \times 10 \quad \text{(Eq. 5)}$$

M is the weight of the dried sample; X_1 is the initial weight of the cup; X_2 is the weight of the cup with oil.

2.3.6. Sensory evaluation test

Sensory evaluation of cheese samples was conducted by 15 evaluators from the Department of Food Science and Engineering, Urmia University, using a 5-point hedonic method for color, odor, taste, mouthfeel, texture, and overall acceptability. The number 1 represented the lowest score and the number 5 the highest score [34].

2.4. Microbial tests of packaged Koupeh cheese samples

2.4.1. Total count of microorganisms

For the total count of microorganisms, 10 gr of each cheese sample was taken and completely crushed under sterile conditions with a sterile mortar and mixed with 90 ml of sodium citrate (2% w/v). In the next step, 1 ml of the resulting suspension was taken and mixed with 9 ml of sterile physiological serum and this was continued until a dilution of 10^{-7} was obtained. For the total count of microorganisms, the Pour Plate culture method was used using nutrient agar culture medium. The culture medium was sterilized in an autoclave and then cooled to 50 °C. In the next step, 1 ml of the last two serial dilutions was poured into sterile 9 cm lined plates, approximately 30 ml of culture medium was poured into it, and stirred according to the standard method. After the culture medium solidified next to the flame, it was placed upside down in an incubator at 37 °C for 48 hours.

Colony counting was performed using a colony counter, and the results were reported as the logarithm of the number of colony units of microorganisms per gram of cheese [35].

2.4.2. Mold and yeast count

For total mold and yeast counts, 10 g of each cheese sample was taken and thoroughly crushed under sterile conditions with a sterile mortar and mixed with 90 ml of sodium citrate (2% w/v). In the next step, 1 ml of the resulting suspension was taken and mixed with 9 ml of sterile physiological serum to obtain a dilution of 0.01. To count mold and yeast, the surface culture method was used using Potato Dextrose Agar (PDA, Scharlau Chemie, Barcelona, Spain). The culture medium was sterilized in an autoclave and then cooled to 50 °C. In the next step, 0.1 ml of the prepared dilutions (0.1 and 0.01) were poured into sterile plates containing solid culture medium to half the height of the plate, then spread evenly with a sterile L-shaped rod and placed upside down in an incubator at 25 °C for 5 days. Colony counting was performed using a colony counter and the results were reported as the logarithm of the number of colony units of microorganisms per gram of cheese [35].

3. Statistical analysis

According to Table (1), two independent variables, storage time and packaging type, were examined at five levels in 15 treatments using a factorial design method. Regression and analysis of variance methods were used to examine the fit of the models, and the type I error level in this study was considered to be 0.05.

Table (1): Statistical scheme of Koupeh cheese packaging

Run	Time (days)	Packaging type
1	60	Blank Film
2	0	Blank Film
3	15	NO Film
4	60	Max MEX/NPs
5	15	Max MEX/NPs
6	60	Optimum Film
7	60	NO Film
8	40	Max MEX/NPs
9	15	Max MEX/NPs
10	40	Optimum Film
11	40	NO Film
12	30	Blank Film
13	15	Optimum Film
14	15	NO Film
15	15	Optimum Film

Blank Film: Control film

Max NPS+EX Film: Film containing the highest amount of graphene oxide nanoparticles (4.0% w/v) and *Silybum marianum* extract microcapsules (15 ml v/v)

Film EX Film: Film containing the highest amount of *Silybum marianum* extract microcapsules (15 ml v/v)

Max NPS Film: Film containing the highest amount of graphene oxide nanoparticles (4.0% w/v):

OPTIMUM Film: Film containing 11.35 ml (v/v) *Silybum marianum* extract microcapsules and 3.41% (w/v) graphene oxide nanoparticles

NO Film: Cheese sample packaging without film

4. Results and discussion

4.1. Results of physicochemical tests of Koupeh cheese samples packaged with nanocomposite films

4.1.1. pH and acidity of Koupeh cheese packaged with nanocomposite films

According to the results of statistical analysis, the effect of time on pH and acidity of Koupeh cheese samples packaged with nanocomposite film of *vicia ervilia protein isolate - Launaea acanthodes* gum containing graphene oxide nanoparticles and encapsulated *Silybum marianum* extract was significant ($P<0.05$). As can be seen in Figures (2 and 3), the pH and acidity of Koupeh cheese samples packaged with nanocomposite film decreased and increased respectively with storage time ($P<0.05$). During 60 days of storage, the lowest pH and highest acidity were related to the cheese samples without packaging. The results showed that during 60 days of storage, the pH decreased significantly and the acidity increased in all tested treatments. The activity of starters and lactic acid-producing bacteria produced lactic acid by fermenting lactose, and as a result, the acidity increased and the pH decreased [36]. However, the least change in pH decrease and the least change in acidity increase were related to the cheese samples in the optimal film and the film containing the maximum variables of graphene oxide nanoparticles and *Silybum marianum* extract microcapsules, which probably helps the phenolic compounds in *Silybum marianum* extract to maintain the pH and acidity of the cheese [34]. The relationship between pH and acidity of cheese is not only dependent on lactic acid produced by microbial flora, but also the buffering capacity of curd, which itself is due to the amount of casein, citrate and phosphate [37]. Similar results were reported by Kavas et al [30] who stated that pH and acidity in Kashar cheese coated with edible films enriched with thyme and clove essential oils decreased and increased, respectively; Ríos-de-Benito et al [38] also reported similar results regarding the decrease in pH and increase in acidity on Panela cheese coated with an edible coating based on chitosan and sodium caseinate containing oregano essential oil and silica nanoparticles. The predictive model for the pH and acidity of the cheese samples is given in equations (6) and (7):

A $pH=4.63-2.50\times Time$ $(Adj\text{-}R^2=93\%)$ (Eq. 6)

$pH=4.65-2.78\times Time$ B

C $pH=4.66-3.08\times Time$

D $pH=4.70-6.70\times Time$

A: Blank Film B: Max MEX/NPs Film C: Optimum Film D: NO Film

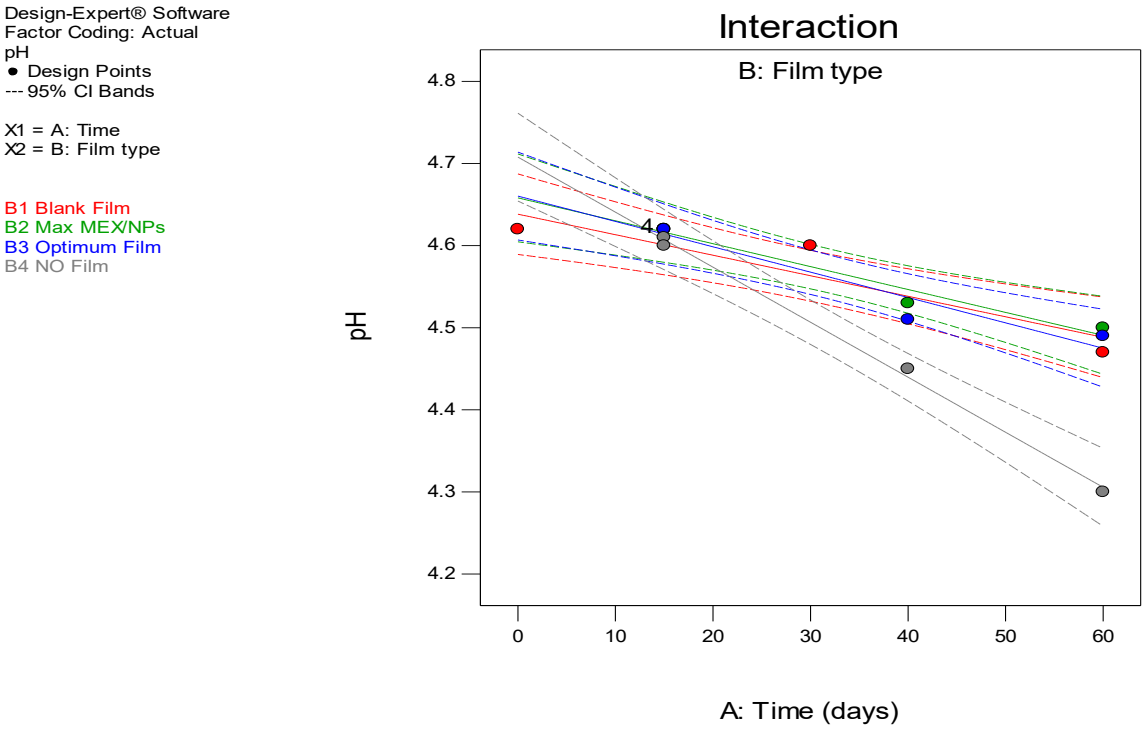

Figure (2): PH interaction of unpackaged and packaged Koupeh cheese with nanocomposite films

A Acidity(%)=0.80+1.33× Time (Adj-R^2=89%) (Eq. 7)

B Acidity(%)=0.78+1.75×Time

C Acidity(%)=0.78+1.75× Time

D Acidity(%)=0.77+3.29× Time

A: Blank Film B: Max MEX/NPs Film C: Optimum Film D: NO Film

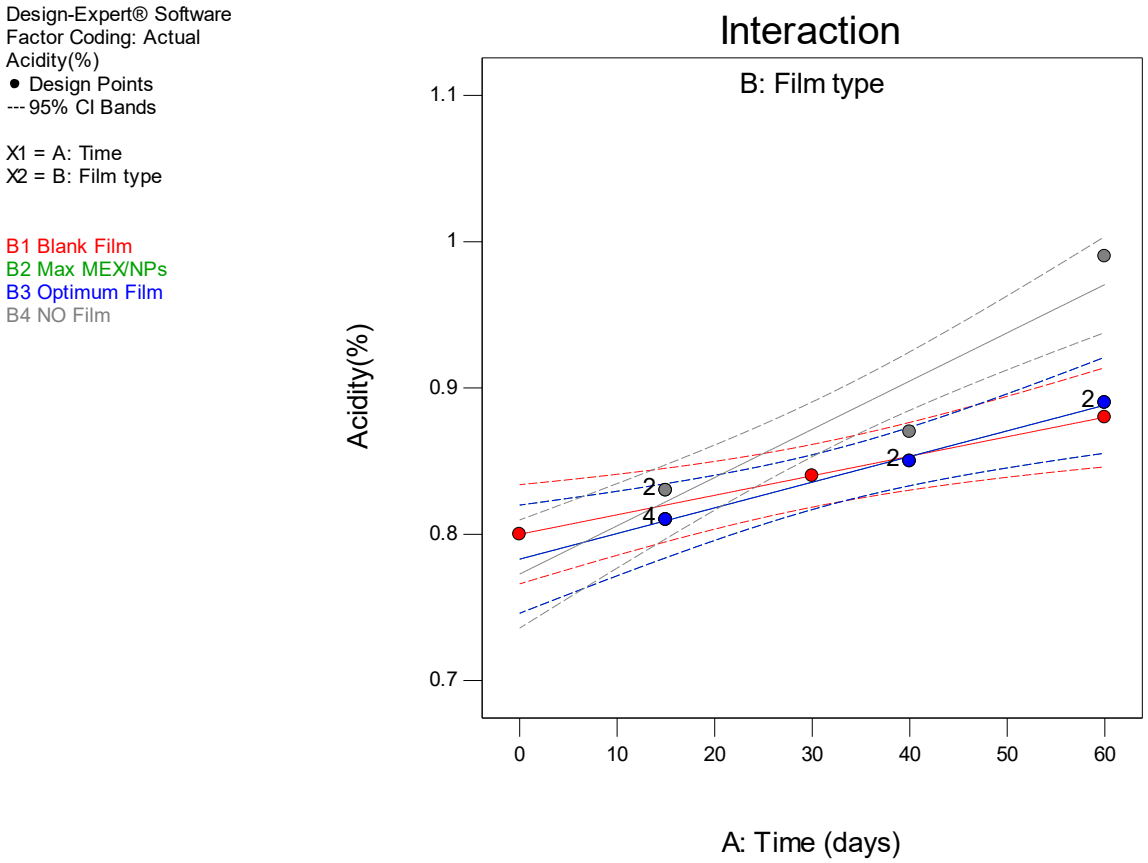

Figure (3): Acidity interaction of unpackaged and packaged Koupeh cheese with nanocomposite films

4.1.2. Salt content of Koupeh cheese packaged with nanocomposite films

The effect of storage time and film type on the salt content of Koupeh cheese samples packaged with *vicia ervilia protein isolate* - *Launaea acanthodes* gum nanocomposite film containing graphene oxide nanoparticles and encapsulated *Silybum marianum* extract was significant (P<0.05). According to Figure (4), the salt content of Koupeh cheese samples packaged with nanocomposite film increased with time and storage time. Salt functions in cheese as a preservative in warm climates and affects the aroma and flavor of cheese and is an important factor in water activity, thus interfering with microbial growth and activity, enzyme activity, reduction in moisture content, biochemical changes during cheese ripening, and simultaneously creating desirable aroma and flavor [36, 39]. The moisture content of cheese affects the rate of salt absorption or diffusion. Also, the rate of salt diffusion in cheese depends on the ratio of fat to solid non-fat (SNF) and moisture to SNF [40]. According to Figure (3), the salt content of Koupeh cheese samples packaged with nanocomposite film increased over time and during storage, but the salt increase rate was low during the first 30 days of storage, and the lowest and highest salt content on the 60th day of cheese storage were, respectively, for Koupeh cheese packaged with nanocomposite film containing maximum variables of graphene oxide nanoparticles and encapsulated *Silybum marianum* extract and the cheese sample without Packaging. Because the polymers forming the film, along with fillers such as nanoparticles, act as a natural barrier and thus reduce the exchange of gas and water between the packaged cheese and the environment [41]. The reason for the increase in salt content during 60 days of storage is probably the low relative humidity of the cheese storage environment and the transfer of moisture from the cheese to the environment, as well as the evaporation of some water from the cheese, which leads to a decrease in moisture and an increase in salt in the dry matter [42]. Also, according to research conducted by Fox et al [43] and Mistry & Atweh et al. [44], the reason for the increase in salt absorption rate can be attributed to the reduction in fat content. For this reason, during 60 days of storage, the

lowest and highest salt content were respectively related to cheese packaged with a nanocomposite film containing maximum variables of graphene oxide nanoparticles and *Silybum marianum* extract microcapsules and the cheese sample without packaging [41]. Khazai et al. [45] reported similar results for Koupeh cheese packaged in biodegradable film based on polyvinyl alcohol and pinto bean starch containing essential oils of garlic, ginger and cinnamon, and El-Sisi et al. [46] reported similar results for cheese coated with chitosan. The predictive model for salt of cheese samples is given in equations (8):

A
Salt(%)=3.59+1.40× Time (Adj-R^2=96%) (Eq. 8)

Salt(%)=3.58+1.40× Time B

Salt(%)=3.59+1.40× Time C

Salt(%)=3.60+1.40×Time D

A: Blank Film B: Max MEX/NPs Film
C: Optimum Film D: NO Film

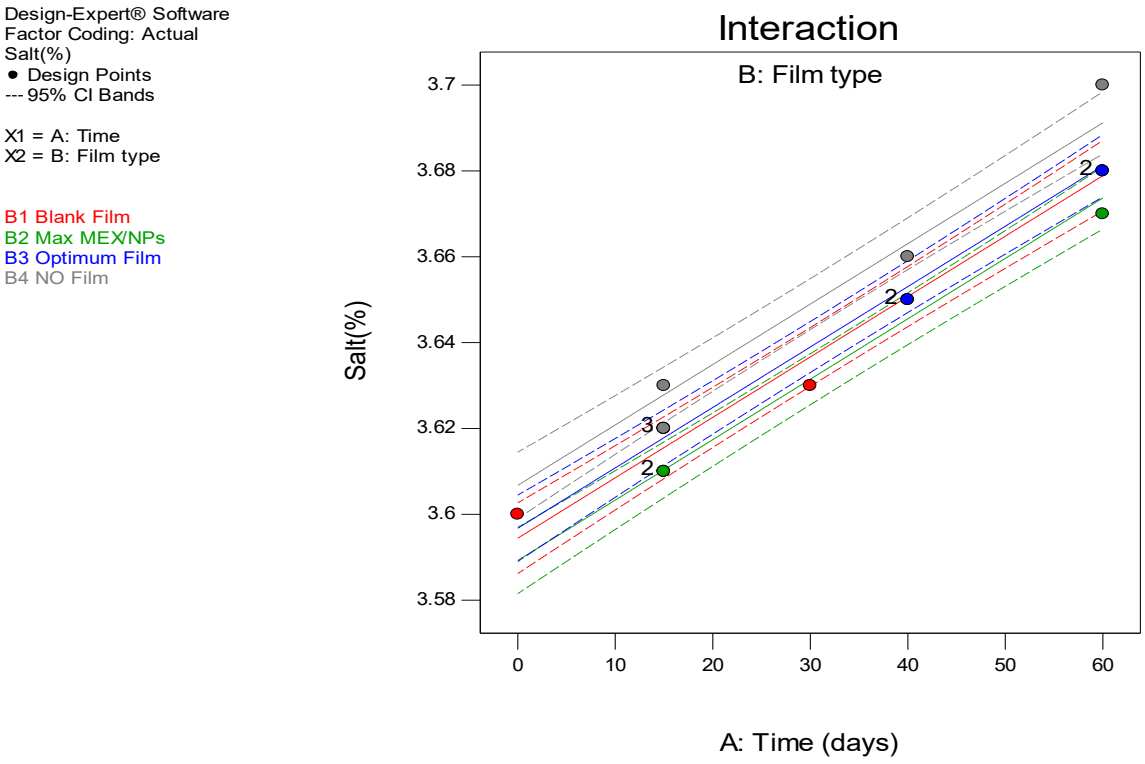

Figure (4): Interaction of unpackaged and packaged Koupeh cheese salt with nanocomposite films

4.1.3. Fat and protein content of Koupeh cheese packaged with nanocomposite films

Regarding the protein content of freshly prepared Koupeh cheese (21.2%), the protein content of packaged and unpackaged cheese samples (21.2%) remained constant during the 60-day storage period without any significant effect of film type (P>0.05). Therefore, film packaging had no effect on the protein content of Koupeh cheese samples. Similar results regarding the lack of effect of film on the protein content of cheese were reported by Nemati et al [47] using whey protein edible film with cumin essential oil in cheese. According to

Figure (5), the fat content of cheese samples decreased significantly over the storage period ($P<0.05$). The fat content of the cheese samples during the first 15 days of storage had a nearly constant trend, and on days 40 and 60 of storage, the highest fat percentage reduction was for the unpackaged cheese sample and the lowest fat percentage reduction was for the Koupeh cheese sample packaged in the Max, Optimum and control films, respectively, and during this storage period, there was a very slight difference between the fat content of the cheese samples packaged in the Maximum and Optimum films. In general, the fat content of the cheese samples in the nanocomposite film decreased more slightly than that of the unpackaged cheese sample, which may be due to the structure of the nanocomposite film and the presence of nanofillers such as nanoparticles causing a smaller decrease in moisture content and, consequently, a smaller increase in dry matter compared to the unpackaged cheese sample [48]. On the other hand, it is possible that the slight decrease in fat content of the cheese samples in the nanocomposite film during the storage period (60 days; $P < 0.05$) may be due to the inhibition of lipolysis in Koupeh cheese. [35] The effective factor in this result could be related to different production conditions, cheese milk fat, type of packaging, presence of pores in the packaging container, etc [49]. Similar results were reported by Bonilla et al. [35] for reducing the fat percentage of cheese packaged with gelatin-chitosan edible film containing Boldo extract. The predictive model for fat of cheese samples is given in equations (9):

(Adj-R^2=95%) (Eq. 9)

A Fat=+25.00-4.16× Time

B Fat=25.03-2.40× Time

C Fat=25.03-2.62× Time

D Fat=25.10-6.42× Time

A: Blank Film B: Max MEX/NPs Film
C: Optimum Film D: NO Film

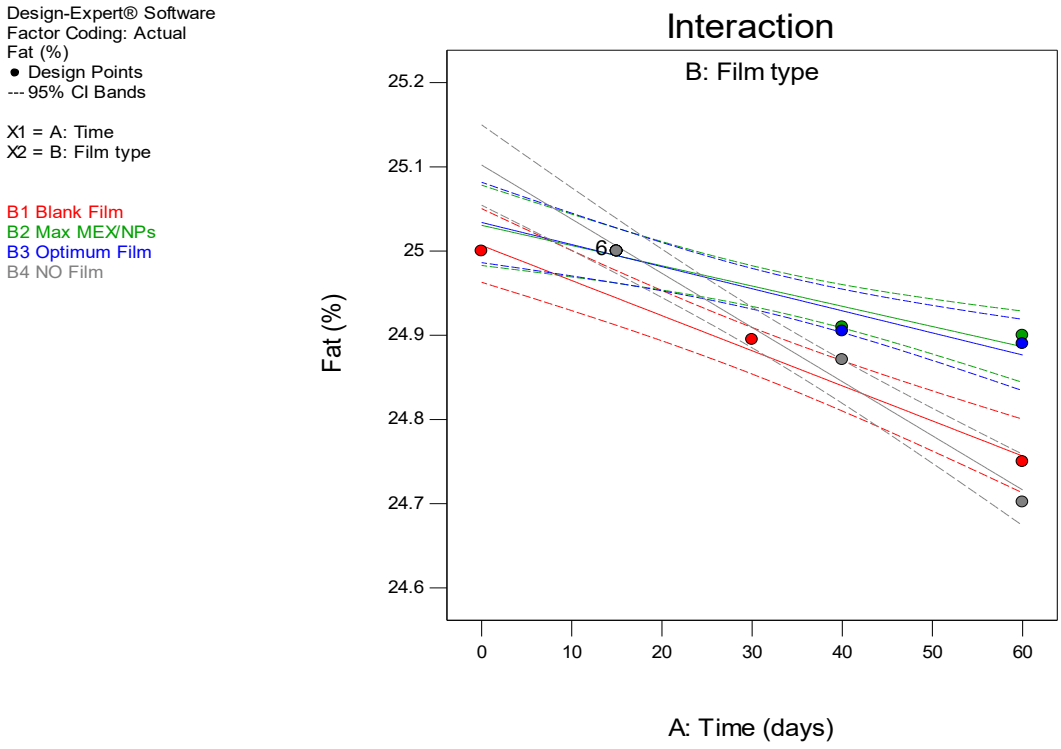

Figure (5): Fat interaction of unpackaged and packaged Koupeh cheese with nanocomposite films

4.1.4. Sensory evaluation of Koupeh cheese packaged with nanocomposite films

Sensory evaluation of Koupeh cheese without packaging and Koupeh cheese packaged in a nanocomposite film based on *vicia ervilia protein isolate - Launaea acanthodes* gum

containing graphene oxide nanoparticles and encapsulated *Silybum marianum* extract and control film during storage time (60 days) is shown in Figure (6). The results showed that increasing storage time and packaging type had no significant effect on the sensory evaluation of the cheese samples (P>0.05).

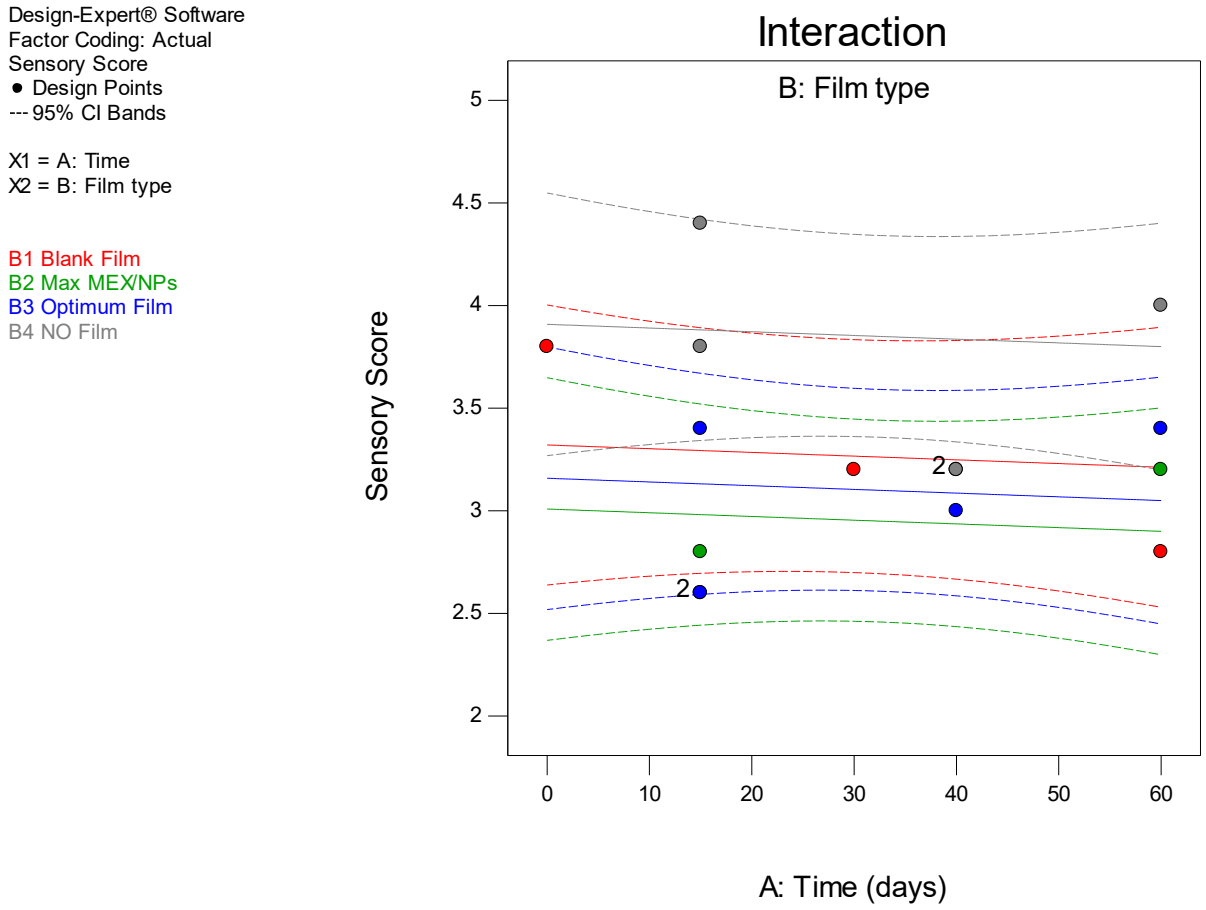

Figure (6): Sensory evaluation interaction of unpackaged and packaged Koupeh cheese with nanocomposite films

4.2. Results of microbiological tests of Koupeh cheese samples packaged with nanocomposite films

4.2.1. Total count of microorganisms in Koupeh cheese samples packaged with nanocomposite films

Microbiological analysis of cheese samples is shown in Figure (7). Microbiological analyses showed statistically significant differences between cheese samples (P<0.05). This study was conducted on unpackaged cheese samples, Koupeh cheese samples packaged with control film, optimal film and film containing maximum variables. As can be seen in Figure (7), the total number of bacteria decreased during the 60-day storage period, but the decrease trend was less during the first 30 days and from the 40th day onwards, a greater decrease in the total number of bacteria was observed. In general, a greater decrease in the total number of bacteria was observed in the Koupeh cheese samples packaged in the film containing the maximum variables, the optimal film, the control film, and finally the Koupeh cheese sample without packaging. The decrease in the total number of microorganisms in the cheese samples may be due to the extract and nanoparticles used in the nanocomposite film, along with the decrease in pH and acid production during the storage period.[45] However, graphene oxide nanoparticles cause chemical oxidation of cellular components, perforation of the bacterial cell wall, and trapping of cells in the graphene oxide structure, which causes bacterial cell death;

also, the oxygen-containing functional groups of graphene oxide destroy the cell membrane through oxidative stress, resulting in the destruction of intracellular materials and metabolic functions of bacterial cells.[25] Also, *Silybum marianum* extract, because it contains phenolic compounds, has antimicrobial properties through cell membrane destruction, changes in the electron transport chain, coagulation of cytoplasmic protein materials, and production of reactive oxygen species (ROS) [50]. Therefore, the total number of microorganisms in the maximum film increased from 3.58 CFU/g to 2.30 CFU/g, in the optimal film from 3.60 CFU/g to 2.50 CFU/g, and in the unpackaged sample from 3.62 CFU/g to 2.95 CFU/g on days 15 and 60 of storage, respectively. Similar results were reported by Mushtaq et al. [51] for freshly packaged Himalayan cheese with zein film containing pomegranate peel extract. Also, Kouser et al. [52] stated that the bioactive edible film based on Aloe barbadensis improves the microbial quality of cheese. The predictive model for the total count of microorganisms in cheese samples is given in equation (10):

A $Log_{10(TC(cfu/g))} = 2.56 - 1.44\times$ Time (Adj-R^2=91 (Eq. 10)

B $Log_{10(TC(cfu/g))} = 2.61 - 4.45\times$ Time

C $Log_{10(TC(cfu/g))} = 2.60 - 3.69\times$ Time

D $Log_{10(TC(cfu/g))} = 2.58 - 1.79\times$ Time

A: Blank Film B: Max MEX/NPs Film
C: Optimum Film D: NO Film

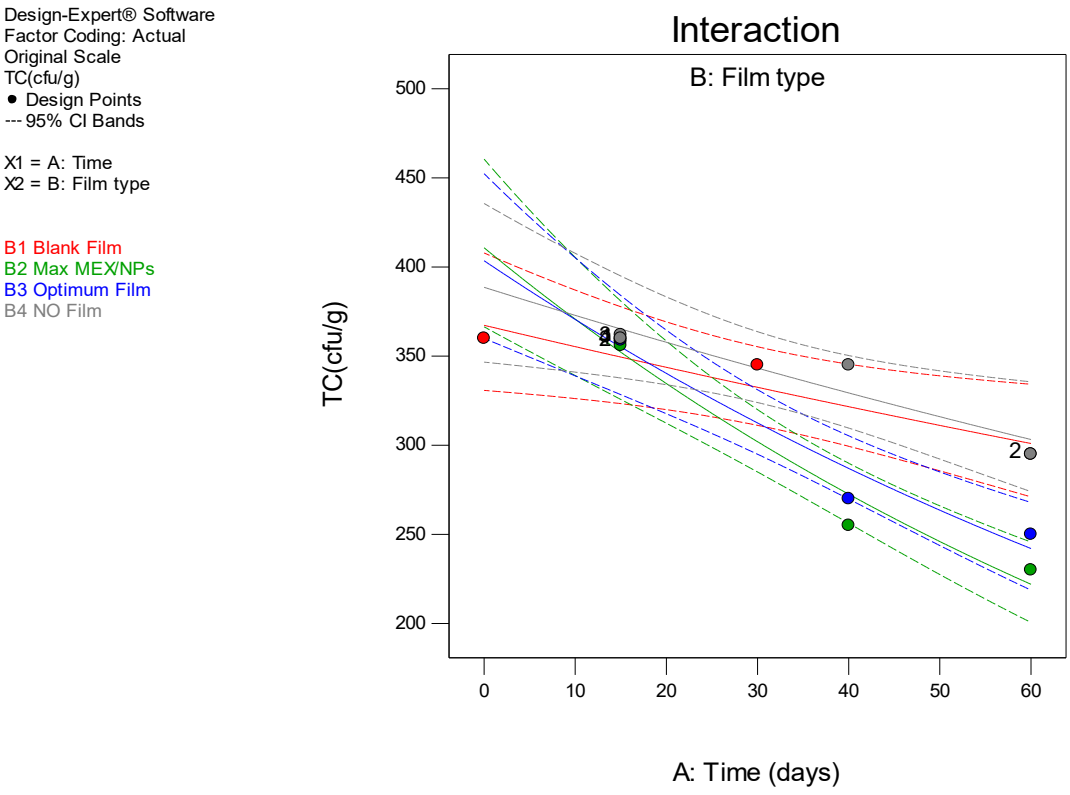

Figure (7): Interaction of the total count of microorganisms of Koupeh cheese without packaging and packaged with nanocomposite films

4.2.2. Mold and yeast counts in samples of Koupeh cheese packaged with nanocomposite films

Figures (8 and 9) show the results of mold and yeast counts of samples of unpackaged Koupeh cheese, Koupeh cheese packaged with Maximum film, optimal film, and control film during a 60-day storage period. In all samples, an increasing trend in the number of mold and yeast was observed during the first 30 days of

storage, and then the amount of mold and yeast decreased significantly ($P<0.05$) in the next 30 days of storage. On day 60 of storage, the mold count was 2 CFU/g for the maximum film, 4 CFU/g for the optimal film, 35 CFU/g for the control film, and 25 CFU/g for the sample without film, and the yeast count was 25 CFU/g for the maximum film, 35 CFU/g for the optimal film, 51 CFU/g for the control film, and 41 CFU/g for the sample without film. These results are probably due to the presence of graphene oxide nanoparticles and *Silybum marianum* extract, which contain phenolic compounds, and these compounds cause antimicrobial properties through cell membrane destruction, changes in the electron transport chain, coagulation of cytoplasmic protein materials, and the production of reactive oxygen species (ROS) [50]. It may also be due to the resistance of these microorganisms to moisture reduction and lactose consumption by lactic acid bacteria and the acidification of the environment (due to their ability to decompose acid), which led to favorable growth of mold and yeast. As a result, an increasing trend of mold and yeast was observed in the cheese samples during the first 30 days of storage. Improper production and storage conditions and lack of hygienic production conditions are factors that increase mold and yeast, which as a result of the high increase of mold and yeast, due to the consumption of lactic acid by molds, causes a decrease in acidity [53]. The maximum permissible limit of mold and yeast in cheese types according to the Iranian National Standard No. 2406 is 102 CFU/g. The amount of mold and yeast in all Koupeh cheese samples packaged in *vicia ervilia protein isolate* - *Launaea acanthodes* gum nanocomposite film containing encapsulated *Silybum marianum* extract and graphene oxide nanoparticles was within the permissible standard range during 60 days of storage in the refrigerator at 4 °C [45]. Similar results were reported by Fajardo et al [54] for the reduction of mold and yeast in Saloio cheese samples packaged with chitosan-based edible film as a carrier for natamycin, Also, Hassas et al. [55] used ethanolic extract of oregano in the formulation of Koupeh cheese and reported that the amount of mold and yeast in the aforementioned samples was significantly reduced compared to the control sample that did not contain the extract. The predictive model for the mold and yeast counts of cheese samples is given in equation (11) and (12), respectively:

A Mold count(cfu/g)= $42.98+0.83\times \text{Time}-0.01\times \text{Time}^2$ (Adj-R^2=92%) (Eq. 11)

B Mold count(cfu/g)= $45.43+0.32\times \text{Time}-0.01\times \text{Time}^2$

C Mold count(cfu/g) =$46.86+0.33\times \text{Time}-0.01\times \text{Time}^2$

D Mold count(cfu/g)= $40.32+0.72\times \text{Time}-0.01\times \text{Time}^2$

A $\text{Log}_{10(\text{Yeast(cfu/g)})}= 1.82+2.65\times \text{Time}-7.71\times \text{Time}^2$ (Adj-R^2=97%) (Eq. 12)

B $\text{Log}_{10(\text{Yeast(cfu/g)})}= 1.90714-3.57\times \text{Time}-7.71\times \text{Time}^2$

C $\text{Log}_{10(\text{Yeast(cfu/g)})}=1.86655-8.44\times \text{Time}-7.71\times \text{Time}^2$

D $\text{Log}_{10(\text{Yeast(cfu/g)})}= 1.84126+5.27\times \text{Time}-7.71\times \text{Time}^2$

A: Blank Film B: Max MEX/NPs Film
C: Optimum Film D: NO Film

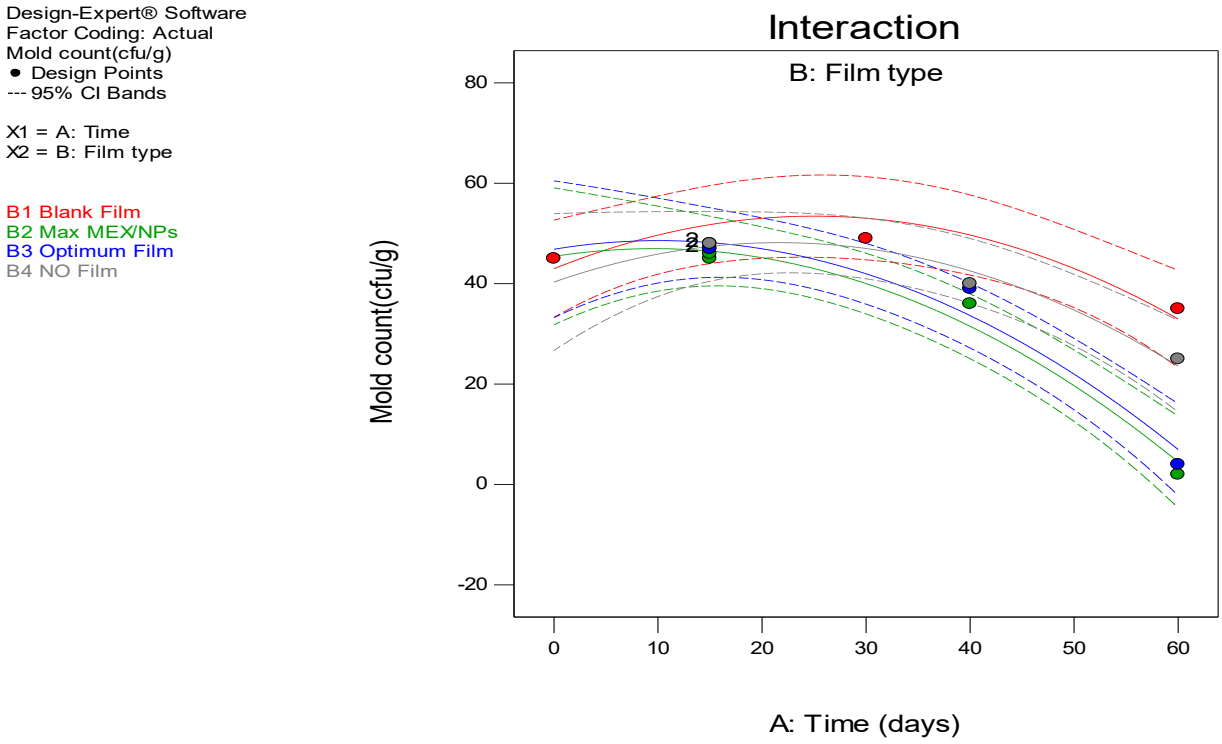

Figure (8): The mold interaction of unpackaged and packaged Koupeh cheese with nanocomposite films

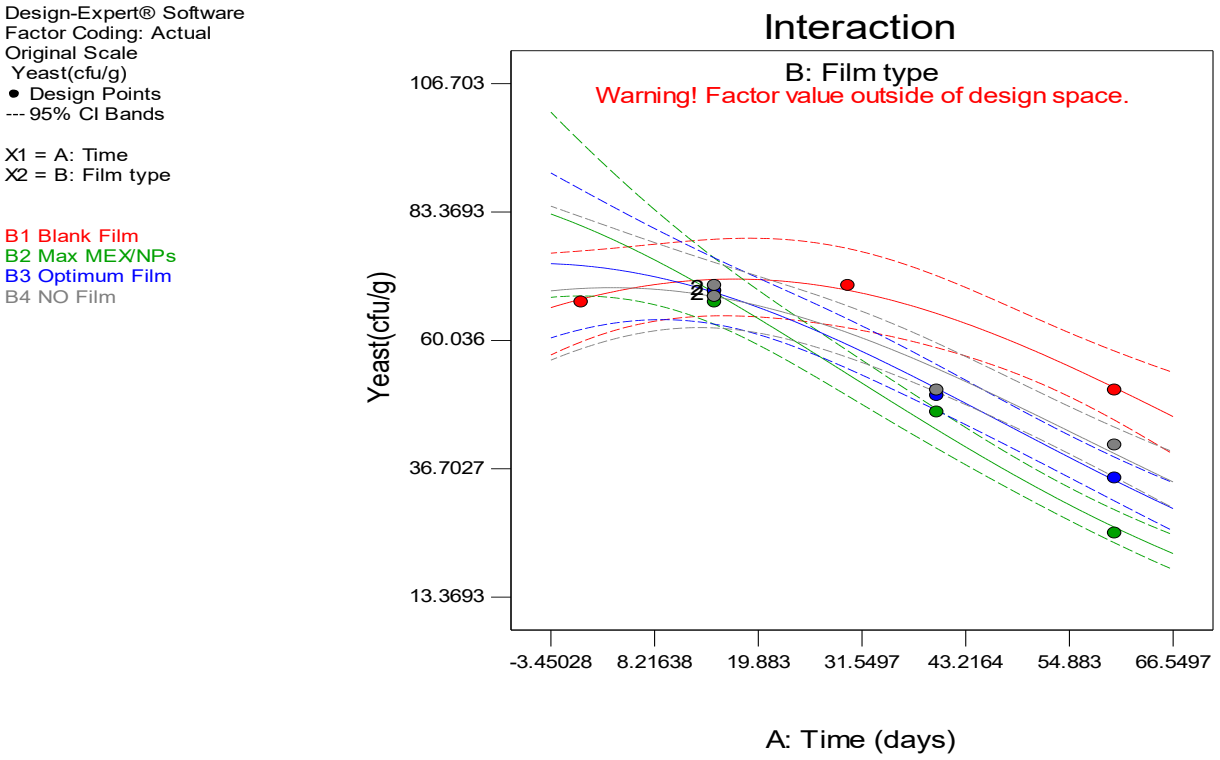

Figure (9): The yeast interaction of Koupeh cheese without packaging and packaged with nanocomposite films

5. Conclusion

In this study, Koupeh cheese was packaged with a nanocomposite film of in *vicia ervilia protein isolate - Launaea acanthodes* gum containing microcapsules of *Silybum marianum* extract and graphene oxide nanoparticles, and the physicochemical and microbial properties of the cheese were investigated during 60 days of storage at 4°C. The effect of film packaging on Koupeh cheese during the storage period caused a significant decrease in pH and fat, a significant increase in acidity and salt, and its values were within the range specified in the Iranian national standard. The results also showed that increasing the storage time and type of packaging had no significant effect on the protein content and sensory evaluation of the cheese samples (P>0.05). The microbial characteristics of the cheese during 60 days of storage were within the permissible range of the national standard. As a result, the physicochemical and microbial properties of Koupeh cheese packaged with a nanocomposite film of in *vicia ervilia protein isolate - Launaea acanthodes* gum containing microcapsules of *Silybum marianum* extract and graphene oxide nanoparticles improved during 60 days of storage.

6.Reference

[1] Hadidi, M., et al., *Plant protein-based food packaging films; recent advances in fabrication, characterization, and applications.* Trends in Food Science & Technology, 2022. **120**: p. 154-173.

[2] Dehnavi, F., A.A. KHOSROWSHAHI, and S. Zomorodi, *Viability of Lactobacillus Acidophilus and its effect on characteristics of Jug cheese.* 2013.

[3] Ghaderi, M., et al., *Isolation and identification of Lactic Acid Bacteria in traditional Pot cheese.* 2013.

[4] Edalatian Dovom, M.R., et al., *Isolation and Identification of the Indigenous Lactic acid Bacteria of Koozeh Cheese and Its Changes during Ripening (Fresh and Ripened) Using Cultural Methods and Carbohydrate Fermentation Profiles.* Iranian Food Science and Technology Research Journal, 2013. **9**(2).

[5] Alizadeh, M., M. Hamedi, and A. Khosroshahi, *Optimizing sensorial quality of Iranian white brine cheese using response surface methodology.* Journal of food science, 2005. **70**(4): p. S299-S303.

[6] Tarahi, M., F. Shahidi, and S. Hedayati, *A Novel starch from bitter vetch*) *Vicia ervilia) seeds: a comparison of its physicochemical, structural, thermal, rheological and pasting properties with conventional starches.* International Journal of Food Science & Technology, 2022. **57**(10): p. 6833-6842.

[7] Arabestani, A., et al., *Properties of a new protein film from bitter vetch (Vicia ervilia) and effect of CaCl2 on its hydrophobicity.* International Journal of Biological Macromolecules, 2013. **57**: p. 118-123.

[8] Porta, R., et al., *Blend films of pectin and bitter vetch (Vicia ervilia) proteins: Properties and effect of transglutaminase.* Innovative Food Science & Emerging Technologies, 2016. **36**: p. 245-251.

[9] Porta, R., et al., *Microstructure and properties of bitter vetch (Vicia ervilia) protein films reinforced by microbial transglutaminase.* Food Hydrocolloids, 2015. **50**: p. 102-107.

[10] Abdolsattari P, Rezazadeh-Bari M, Pirsa S. *Smart film based on polylactic acid, modified with polyaniline/ZnO/CuO: Investigation of physicochemical properties and its use of intelligent packaging of orange juice*. Food and Bioprocess Technology. 2022, **15**(12):2803-25.

[11] Piazza, L., S. Bertini, and J. Milany, *Extraction and structural characterization of the polysaccharide fraction of Launaea acanthodes gum.* Carbohydrate polymers, 2010. **79**(2): p. 449-454.

[12] Ghaderian, S. and A. Baker, *Geobotanical and biogeochemical reconnaissance of the ultramafics of Central Iran.* Journal of Geochemical Exploration, 2007. **92**(1): p. 34-42.

[13] Nergard, C.S., et al., *Isolation, partial characterisation and immunomodulating activities of polysaccharides from Vernonia kotschyana Sch. Bip. ex Walp.* Journal of Ethnopharmacology, 2004. **91**(1): p. 141-152.

[14] Amiri S, Roshani Saray F, Rezazad-Bari L, Pirsa S. *Optimization of extraction and characterization of physicochemical, structural, thermal, and antioxidant properties of mucilage from Hollyhock's root: a functional heteropolysaccharide*. Journal of Food Measurement and Characterization. 2021, **15**:2889-903.

[15] Sahi, S., H. Djidjelli, and A. Boukerrou, *Study of the properties and biodegradability of the native and plasticized corn flour-filled low density polyethylene composites for food packaging applications.* Materials Today: Proceedings, 2021. **36**: p. 67-73.

[16] Rahimi, A. and M. Kamali, *Different planting date and fertilizing system effects on the seed yield, essential oil and nutrition uptake of milk thistle (Silybum marianum (L.) Gaertn.).* Advances in Environmental Biology, 2012. **6**(5): p. 1789-1796.

[17] Fallah Huseini, H., A. Hemati, and S. Alavian, *A review of herbal medicine: Silybum marianum.* Journal of Medicinal Plants, 2004. **3**(11): p. 14-24.

[18] Tooiserkani, F., A. Hormati, and H. Moradi, *A Glimpse of Silybum marianum from the Perspective of Iranian Traditional Medicine and Modern Studies.* Qom University of Medical Sciences Journal, 2019. **13**(1): p. 78-86.
[19] Lucini, L., et al., *Phenolic profile and in vitro antioxidant power of different milk thistle [Silybum marianum (L.) Gaertn.] cultivars.* Industrial Crops and Products, 2016. **83**: p. 11-16.
[20] Serçe, A., et al., *Assessment of the antioxidant activity of Silybum marianum seed extract and its protective effect against DNA oxidation, protein damage and lipid peroxidation.* Food technology and biotechnology, 2016. **54**(4): p. 455-461.
[21] Tang, Y., et al., *Electrospun gelatin nanofibers encapsulated with peppermint and chamomile essential oils as potential edible packaging.* Journal of agricultural and food chemistry, 2 :(٨)٦٧ .٠١٩p. 2227-2234.
[22] Gautam, S., et al., *Antibacterial efficacy of poly (vinyl alcohol) nanocomposites reinforced with graphene oxide and silver nanoparticles for packaging applications.* Polymer Composites, 2021. **42**(6): p. 2829-2837.
[23] Xie, Y.-Y., et al., *Development and antibacterial activities of bacterial cellulose/graphene oxide-CuO nanocomposite films.* Carbohydrate Polymers, 2020. **229**: p. 115456.
[24] .٢٤ Usman, A., et al., *Enhanced mechanical, thermal and antimicrobial properties of poly (vinyl alcohol)/graphene oxide/starch/silver nanocomposites films.* Carbohydrate polymers, 2016. **153**: p. 592-599.
[25] Das, M., et al., *Synergetic reinforcing effect of graphene oxide and nanosilver on carboxymethyl cellulose/sodium alginate nanocomposite films: Assessment of physicochemical and antibacterial properties.* International Journal of Biological Macromolecules, 2023. **239**: p. 124185.
[26] Ghasemizad S, Pirsa S, Amiri S, Abdosatari P. *Optimization and characterization of bioactive biocomposite film based on orange peel incorporated with gum arabic reinforced by Cr2O3 nanoparticles*. Journal of Polymers and the Environment. 2022, **30**(6):2493-506.
[27] Pirsa S, Bener M, Şen FB. *Biodegradable film of carboxymethyl cellulose modified with red onion peel powder waste and boron nitride nanoparticles: Investigation of physicochemical properties and release of active substances*. Food Chemistry. 2024, **1**;445:138721.
[28] Pirsa S, Mahmudi M, Ehsani A. *Biodegradable film based on cress seed mucilage, modified with lutein, maltodextrin and alumina nanoparticles: Physicochemical properties and lutein controlled release*. International Journal of Biological Macromolecules. 2023, **1**;224:1588-99.
[29] Alizadeh, M., *Packaging of UF cheese with edible film of nanocomposite isolated mung bean protein-apple pectin containing microencapsulation of cardamom extract and serum oxide nanoparticles and quantum carbon Graphite dots: Investigation of its physicochemical properties.* Journal of food science and technology (Iran), 202 :(١٢٨)١٩ .٢p. 235-247.
[30] Kavas, G., N. Kavas, and D. Saygili, *The effects of thyme and clove essential oil fortified edible films on the physical, chemical and microbiological characteristics of kashar cheese.* Journal of food quality, 2015. **38**(6): p. 4.٤١٢-٠٥
[31] Momeni Sarvestani, M. and H. Lashkari, *Effect of black cumin essential oil on physicochemical, microbial and sensorial characteristics of ultrafilterated Feta cheese.* Iranian Food Science and Technology Research Journal, 2020. **16**(4): p. 409-42.١
[32] Amiri S, Kohneshahri SR, Nabizadeh F. *The effect of unit operation and adjunct probiotic culture on physicochemical, biochemical, and textural properties of Dutch Edam cheese*. LWT. 2022, **1**;155:112859.
[33] Pirsa S, Abdolsattari P, Peighambardoust SJ, Fasihnia SH, Peighambardoust SH. *Investigating microbial properties of traditional Iranian white cheese packed in active LDPE films incorporating metallic and organoclay nanoparticles*. Chemical Review and Letters. 2020, **1**;3(4):168-74.Karunamay, S., S.R. Badhe, and V. Shulka, *Effect of Edible Packaging Film Treated with Essential Oil of Clove in Extending the Shelf Life of Paneer.* 2.٠٢٠
[34] Pirsa S, Dalili R, Yazdani I. *Effects of quince seed mucilage and guar gum on the physicochemical and sensory properties of Doogh*. Journal of Agricultural Science and Technology. 2018, **20**(3):485-94.
[35] Motelica, L., et al., *Antibacterial biodegradable films based on alginate with silver nanoparticles and lemongrass essential oil–innovative packaging for cheese.* Nanomaterials, 2021. **11**(9): p. 2377.
[36] Macedo, A.C., F.X. Malcata, and J.C. Oliveira, *Effect of production factors and ripening conditions on the characteristics of Serra cheese.* International journal of food science & technology, 1997. **32**(6): p. 501-511.
[37] Ríos-de-Benito, L.F., et al., *Design of an active edible coating based on sodium caseinate, chitosan and oregano essential oil reinforced with silica particles and its application on panela cheese.* Coatings, 2021. **11**(10): p. 1212.
[38] Kaplan, N.M., *The dietary guideline for sodium: should we shake it up? No.* The American journal of clinical nutrition, 2000. **71**(5): p. 1020-1026.

[39] Geurts, T.J., P. Walstra, and H. Mulder, *Brine composition and the prevention of the defect'soft rind'in cheese.* 1973.
[40] Mahcene, Z., et al., *Home-made cheese preservation using sodium alginate based on edible film incorporating essential oils.* Journal of food science and technology, 2021. **58**: p. 2406-2419.
[41] Cooke, D., A. Khosrowshahi, and P. Mcsweeney, *Effect of gum tragacanth on the rheological and functional properties of full-fat and half-fat Cheddar cheese.* Dairy Science & Technology, 2013. **93**(1): p. 45-62.
[42] Fox, P.F., et al., *Fundamentals of cheese science*. Vol. 1. 2017: Springer.
[43] Mistry, S.J. and G.F. Atweh, *Stathmin inhibition enhances okadaic acid-induced mitotic arrest: a potential role for stathmin in mitotic exit.* Journal of Biological Chemistry, 2001. **276**(33): p. 31209-31215.
[44] Khazaei, A., L. Nateghi, and N. Zand, *Evaluation of Microbial and Sensory Properties of Jug Cheese Packed in the Biodegradable Film of Polyvinyl Alcohol and Pinto Bean Starch-Containing Garlic, Ginger, and Cinnamon Essential Oils.* Iran. J. Chem. Chem. Eng. Research Article Vol, 2022. **41**(11).
[45] El-Sisi, A.S., A. Gapr, and K. Kamaly, *Use of chitosan as an edible coating in RAS cheese.* Biolife, 2015. **3**(2): p..٥٧٠-٥٦٤
[46] Nemati, V., et al., *Application of a Whey Protein Edible Film Incorporated with Cumin Essential Oil in Cheese Preservation.* Coatings, 2023. **13**(8): p. 1470.
[47] Hayaloglu, A., M. Guven, and P. Fox, *Microbiological, biochemical and technological properties of Turkish White cheese 'Beyaz Peynir'.* International Dairy Journal, 2002. **12**(8): p. 635-648.
[48] Ceylan, Z., H. Turkoglu, and K. Dayisoylu, *The microbiological and chemical quality of sikma cheese produced in Turkey.* pakistan Journal of Nutrition, 2003. **2**(2): p. 95-97.
[49] Gohargani, M., H. Lashkari, and A. Shirazinejad, *Study on biodegradable chitosan-whey protein-based film containing bionanocomposite TiO2 and Zataria multiflora essential oil.* Journal of Food Quality, 2020. **2020**: p. 1-11.
[50] Mushtaq, M., et al., *Use of pomegranate peel extract incorporated zein film with improved properties for prolonged shelf life of fresh Himalayan cheese (Kalari/kradi).* Innovative Food Science & Emerging Technologies, 2018. **48**: p. 25-32.
[51] Kouser, F ,.et al., *Aloe barbadensis based bioactive edible film improved lipid stability and microbial quality of the cheese.* Foods, 2023. **12**(2): p. 229.
[52] Nateghi, *Investigation of the effect of essential oil of the sap of the betel tree on the physicochemical, microbial and sensory properties of jarred cheese.* Food Industry Research, 2021. **31**(2): p. 67-87.[In persian]
[53] Fajardo, P., et al., *Evaluation of a chitosan-based edible film as carrier of natamycin to improve the storability of Saloio cheese.* Journal of food engineering, 2010. **101**(4): p. 349-356.
[54] Hassas, B., L. Nateghi, and A .Lavasani, *Investigation of physicochemical, microbial and sensory properties of low-fat jug cheeses containing β-glucan powder and ethanolic extract of menthe longifolia.* Iranian Food Science & Technology Research Journal, 2019. **15**(1): p. 181-198.

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چکیده

پنیر به دلیل ویژگی‌ها و تنوع زیاد، به‌عنوان چالش‌برانگیزترین فرآورده لبنی شناخته می‌شود که باعث می‌شود پوشش‌ها و فیلم‌های خوراکی به طور گسترده‌ای برای افزایش عمر مفید آن استفاده شوند. بنابراین این پژوهش با هدف کاربرد فیلم نانوکامپوزیت ایزوله پروتئین گاودانه – صمغ چرخک حاوی میکروکپسول‌های عصاره خارمریم در سطوح متفاوت(۰ و ۱۵ حجمی/ حجمی) و نانو ذرات اکسیدگرافن در سطوح متفاوت(۰ و ۰/۴ وزنی/حجمی) جهت بسته‌بندی پنیر کوپه انجام شد. دو فاکتور زمان نگهداری و نمونه-فیلم‌های تولید شده (فیلم شاهد، فیلم حاوی ماکسیمم مقادیر نانوذره و میکروکپسول عصاره خارمریم و فیلم بهینه حاوی ۳/۴۱٪ وزنی/حجمی نانوذره و ۱۱/۳۵ حجمی/حجمی میکروکپسول عصاره خارمریم) در بسته‌بندی پنیر کوپه و نمونه پنیر کوپه بدون پوشش فیلم طبق روش سطح پاسخ طرح فاکتوریل مورد بررسی قرار گرفتند. ویژگی‌های فیزیکوشیمایی(pH، اسیدیته، نمک، چربی، پروتئین)، میکروبی (شمارش کلی میکروارگانیسم‌ها، کپک و مخمر) و ارزیابی حسی پنیر در طی ۶۰ روز نگهداری بررسی شد. تاثیر بسته‌بندی فیلم روی پنیر کوپه طی دوره نگهداری، باعث کاهش pH و چربی، افزایش اسیدیته و نمک شد. همچنین نتایج نشان داد که افزایش زمان نگهداری و نوع بسته‌بندی تاثیر معنی‌داری بر میزان پروتئین و ارزیابی حسی نمونه‌های پنیر نداشت ($P>0.05$). ویژگی‌های میکروبی پنیر طی ۶۰ روز نگهداری در محدوده مجاز استاندارد ملی بود. به طور کلی، فیلم‌های نانوکامپوزیتی ایزوله پروتئین گاودانه-صمغ چرخک حاوی میکروکپسول‌های عصاره خارمریم و نانوذره اکسیدگرافن ممکن است برای استفاده به عنوان مواد بسته‌بندی سازگار با محیط زیست در صنایع غذایی مناسب باشند و همچنین ایمنی محصولات غذایی را در زمان نگهداری افزایش دهند.